

CLAIMS

1. A method for determining the concentration of circulating DNA in a plasma sample from a cancer patient, a subject with cancer susceptibility or at risk of developing cancer, which comprises:

- 1) extracting the DNA from the plasma sample;
- 2) adding to the DNA preparation: a) a mixture of oligonucleotide primers suitable for PCR amplification of a fragment of the human telomerase reverse transcriptase (hTERT) gene, and b) an oligonucleotide probe, having at least one quencher and one reporter fluorophore at the 3' and 5' ends, able to anneal to a sequence within the region delimited by the primers, in suitable conditions for carrying out a PCR reaction,
- 3) adding a heat-stable DNA polymerase with 5'-3' hexonuclease activity and amplifying the hTERT gene fragment;
- 4) measuring the produced fluorescence.

2. A method according to claim 1, wherein the DNA concentration in the test sample is determined by interpolation of a calibration curve calculated with known amounts of DNA.

3. A method as claimed in claims 1-2, which further comprises comparing the concentration of circulating DNA to a reference concentration.

4. A method according to claim 3, wherein the reference concentration is from 9 to 25 ng/ml.

5. A method as claimed in claim 1, wherein said fragment of the human telomerase reverse transcriptase (hTERT) gene is from nt 13059 to nt 13156 of the sequence GenBank accession n. AF128893.

6. A method according to claim 5, wherein said fragment of the human telomerase reverse transcriptase (hTERT) gene is amplified using SEQ ID N.

1 and 2 as the primers forward and reverse, respectively, and SEQ ID N. 3 as the probe.

7. A method as claimed in claims 1-3, for the early diagnosis, prognosis or clinical monitoring of cancer patients.

5 8. A method as claimed in claims 1-3, for the evaluation of the risk of developing cancer in healthy individuals or individuals with familiar cancer susceptibility.

9. A method as claimed in claim 8, for the evaluation of the risk of cancer development in smokers.

10 10. A method as claimed in claim 1, wherein said cancer is lung, colon-rectum, head and neck, liver or pancreas cancer.

11. A method as claimed in claim 10, wherein said cancer is lung carcinoma.